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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.
09/116,502	07/16/98	FALLON R	CL-1035

HM12/0330

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EXAMINER	
FRONDA, C	
ART UNIT	**PAPER NUMBER**
1652	

DATE MAILED: 03/30/00

Please find below and/or attached an Office communication concerning this application or proceeding.

Commissioner of Patents and Trademarks

Office Action Summary	Application No. 09/116,502	Applicant(s) Fallon et al.
	Examiner Christian L. Fronda	Group Art Unit 1652

☐ Responsive to communication(s) filed on _____.

☒ This action is **FINAL.**

☐ Since this application is in condition for allowance except for formal matters, **prosecution as to the merits is closed** in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11; 453 O.G. 213.

A shortened statutory period for response to this action is set to expire 3 month(s), or thirty days, whichever is longer, from the mailing date of this communication. Failure to respond within the period for response will cause the application to become abandoned. (35 U.S.C. § 133). Extensions of time may be obtained under the provisions of 37 CFR 1.136(a).

Disposition of Claims

☒ Claim(s) *1-27* is/are pending in the application.

Of the above, claim(s) *24* is/are withdrawn from consideration.

☒ Claim(s) *2, 7, 18, and 22* is/are allowed.

☒ Claim(s) *1, 3-6, 8-17, 19-21, 23, and 25-27* is/are rejected.

☒ Claim(s) *1* is/are objected to.

☐ Claims _____ are subject to restriction or election requirement.

Application Papers

☐ See the attached Notice of Draftsperson's Patent Drawing Review, PTO-948.

☐ The drawing(s) filed on _____ is/are objected to by the Examiner.

☐ The proposed drawing correction, filed on _____ is ☐approved ☐disapproved.

☐ The specification is objected to by the Examiner.

☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. § 119

☐ Acknowledgement is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d).

☐ All ☐ Some* ☐ None of the CERTIFIED copies of the priority documents have been

☐ received.

☐ received in Application No. (Series Code/Serial Number) _____.

☐ received in this national stage application from the International Bureau (PCT Rule 17.2(a)).

*Certified copies not received: _____.

☒ Acknowledgement is made of a claim for domestic priority under 35 U.S.C. § 119(e).

Attachment(s)

☒ Notice of References Cited, PTO-892

☐ Information Disclosure Statement(s), PTO-1449, Paper No(s). _____

☐ Interview Summary, PTO-413

☐ Notice of Draftsperson's Patent Drawing Review, PTO-948

☐ Notice of Informal Patent Application, PTO-152

--- SEE OFFICE ACTION ON THE FOLLOWING PAGES ---

DETAILED ACTION

Response to Amendment

1. Applicant's amendment, filed January 4, 2000, has been entered and considered. The amendment cancels claim 24 without prejudice in favor of claim 25, amends claims 1, 3-6, 8-13, 16, 20, 21, and 26, and amends the specification.

Claims 2, 7, 18, and 22 were mistakenly rejected in the in the non-final Office Action, Paper 9, mailed August 4, 1999. The rejections on claims 2, 7, 18, and 22 are withdrawn. Claims 2, 7, 18, and 22 are allowed since these claims are novel and unobvious.

Because claims 1, 3-6, 8-13, 16, 20, 21, and 26 have been amended, it is necessary to state new claim rejections under U.S.C. 102 and U.S.C. 103 which are listed below.

Claim Objections

2. Claim 1 objected to because of the following informalities: claim 1 improperly depends on claim 3. Claim 3 should be renumbered as **claim 1**, claim 1 should be renumbered as **claim 2**, and claim 2 should be renumbered as **claim 3**. Appropriate correction is required.

Claim Rejections - 35 U.S.C. § 102

3. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless --
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

Claim 16 is rejected under 35 U.S.C. 102(b) as being anticipated by Masuda *et al.* (1995). Masuda *et al.* (1995) teach *Candida maltosa* mutant strains having no more than both POX4 genes disrupted (see **lines 15-24 of (c) Disruption of POX genes**, p159). In addition **Table 1** (see p. 159) shows that the P4DD strain which has both POX4 genes disrupted cannot use an alkane, alcohol, or a fatty-acid for growth. Therefore, the beta-oxidation pathway has been blocked in strain P4DD.

Claim Rejections - 35 U.S.C. § 103

4. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(f) or (g) prior art under 35 U.S.C. 103(a).

Claims 3-6 are rejected under 35 U.S.C. 103(a) as being unpatentable over Cregg *et al.* in view of Takagi *et al.*, Takagi, Ohkuma, and Ohkuma *et al.* Cregg *et al.* teach how to transform and express foreign genes in *Pichia pastoris* (see entire publication), suitable vectors containing the regulatory element *AOX1* promoter (see **Fig.2**, p. 907), and heterologous proteins produced in *Pichia pastoris* (see **Table 1**, p. 905). Furthermore, Cregg *et al.* teach the advantages of using *Pichia pastoris* as a host cell (see **Conclusion**, p. 909) : the *AOX1* promoter which regulates heterologous protein expression; well-developed methods for genetic manipulation of *Pichia pastoris*; and methods for growth of expression strains in large, high-density fermentor cultures. Cregg *et al.* does not teach a *Pichia pastoris* transformed with *Candida maltosa* genes for cytochrome P450 monooxygenase Alk1-A or cytochrome P450 monooxygenase Alk3-A or cytochrome P450 reductase. Takagi *et al.* teach a *Candida maltosa* gene for cytochrome P450alk (see **Fig.3**, p.2221 and Accession No. D00481). Takagi teach *Candida maltosa* Alk2-A and Alk3-A genes (Accession No.X55881). Ohkuma teach a *Candida maltosa* genes for cytochrome P450 reductase (Accession No. D23327), ALK4 (Accession No. D12716), ALK5-A (Accession No. D12717), ALK6-A (Accession No. D12718), and ALK7 and ALK8 (Accession No. D12719). Ohkuma *et al.* teach *Candida maltosa* genes for ALK2-A and ALK3-A (Accession NO. X55881).

In claims 4 and 5:

A. SEQ ID NO: 35 is anticipated by Takagi *et al.* (Accession No. D00481).

B. SEQ ID NO: 36 is anticipated by Takagi (Accession No. X55881).

C. SEQ ID NO: 37 is anticipated by Takagi (Accession No. X55881).

D. SEQ ID NO: 38 is anticipated by Ohkuma (Accession No. D12716).
E. SEQ ID NO: 39 is anticipated by Ohkuma (Accession No. D12717).
F. SEQ ID NO: 40 is anticipated by Ohkuma (Accession No. D12718).
G. SEQ ID NO: 41 is anticipated by Ohkuma (Accession No. D12719).
H. SEQ ID NO: 42 is anticipated by Ohkuma (Accession No. D12719).
I. SEQ ID NO: 43 is anticipated by Ohkuma (Accession No. D25327).

In claim 6 it is noted that the recited SEQ ID NOs: 35 and 37 are from *Candida maltosa* ATCC 90677. However, SEQ ID NO: 35 is anticipated by Takagi *et al.* (Accession No. D00481) and SEQ ID NO: 37 is anticipated by Takagi (Accession NO.X55881). Therefore, the recited DNA sequences from *Candida maltosa* strain ATCC 90677 are not patentably distinct from D00481 and X55881.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to make a transformed *Pichia pastoris* strain according to claims 3-6 since the rationale, reagents, and process steps for making this strain are known. A transformed *Pichia pastoris* strain according to claims 3-6 is expected to be made by modifying the teachings of Cregg *et al.* as follows:

A. Clone the *Candida maltosa* genes taught by Takagi *et al.*, Takagi, Ohkuma, and Ohkuma *et al* into vectors suitable for *Pichia pastoris* as described by Cregg *et al.* (see **Fig. 2**, p. 907).
B. Transform *Pichia pastoris* as described by Cregg *et al.* with vectors containing the *Candida maltosa* genes for cytochrome P450 monooxygenase Alk1-A or cytochrome P450 monooxygenase Alk3-A or cytochrome P450 reductase.

One of ordinary skill in the art would be motivated to make a transformed *Pichia pastoris* strain according to claims 3-6 because of the advantages of expressing foreign genes in *Pichia pastoris* taught by Cregg *et al.*: the *AOX1* promoter which regulates heterologous protein expression; well-developed methods for genetic manipulation of *Pichia pastoris*; and methods for growth of expression strains in large, high-density fermentor cultures. Furthermore, one of ordinary skill in the art would have had a reasonable expectation for success of making a transformed *Pichia pastoris* strain according to claims 3-6 because of the success of expressing heterologous proteins in *Pichia pastoris* which is reported by Cregg *et al.* (see **Table 1**, p. 905).

Claim 1 is rejected under 35 U.S.C. 103(a) as being unpatentable over Hammond in view of Takagi *et al.*, Takagi, Ohkuma, Ohkuma *et al*, and Picataggio *et al.*(1992). Hammond teaches the bioproduction alkyl dicarboxylic acids from alkyl precursors using immobilized *Candida maltosa* cells (see entire patent). Hammond does not teach bioproduction of C_6 to C_{22} mono- and di-carboxylic acids using a transformed *Pichia pastoris* according to claim 1. The teachings of Takagi *et al.*, Takagi, Ohkuma, and Ohkuma *et al.* are described above. Picataggio *et al.* (1992)

teach the disadvantages of using chemical synthesis to prepare aliphatic long-chain dicarboxylic acids which include the presence of byproducts during synthesis and extensive purification procedures to remove these byproducts (see first paragraph of the **Introduction**, p. 894).

It would have been obvious to one of ordinary skill in the art at the time the invention was made to produce mono- and di-carboxylic acids according to claim 1 since the rationale, reagents, and process steps are known. C_6 to C_{22} mono- and di-carboxylic acids are expected to be made according to the method of claim 1 by modifying the teachings of Hammond in the following manner:

A. Prepare a transformed *Pichia pastoris* which is described above in the 35 U.S.C. 103(a) rejection claims 3-6.

B. Use the genetically-engineered *Pichia pastoris* in the process described by Hammond to produce any mono- or di-carboxylic acid.

One of ordinary skill in the art would be motivated to produce C_6 to C_{22} mono- and di-carboxylic acids according to claim 1 since Picataggio *et al.*(1992) teach the disadvantages of using chemical synthesis to prepare aliphatic long-chain dicarboxylic acids. In addition, the use and advantages of yeasts in the bioproduction of mono- and di-carboxylic acids are well known in the art (for example see US 4, 275, 158 and US 4, 220, 720). Furthermore, one of ordinary skill in the art would have had a reasonable expectation for success in producing mono- or dicarboylic acids according to claim 1 because of the success of Hammond in producing alkyl dicarboxylic acids from alkyl precursors.

Claims 10-13, and 20-22 are rejected under 35 U.S.C. 103(a) as being unpatentable over Kasuske *et al.* in view of Takagi *et al.*, Takagi, Ohkuma, and Ohkuma *et al.* Kasuske *et al.* teach an integrative cloning vector for *Candida maltosa* and methods of transforming *Candida maltosa* with this vector (see **Fig.1**, p. 692 and **MATERIALS AND METHODS** and **RESULTS**, pp.691-697). Kasuske *et al.* do not teach a transformed *Candida maltosa* according to claims 10 and 11. Takagi *et al.*, Takagi, Ohkuma, and Ohkuma *et al.* teach DNA sequences as described above which anticipate SEQ ID NOs: 35-43 recited in claims 11, 13, 20, and 21.

In claim 13 it is noted that the recited SEQ ID NOs: 35 and 37 are from *Candida maltosa* ATCC 90677. However, SEQ ID NO: 35 is anticipated by Takagi *et al.* (Accession No. D00481) and SEQ ID NO: 37 is anticipated by Takagi (Accession NO.X55881). Therefore, the recited DNA sequences from *Candida maltosa* strain ATCC 90677 are not patentably distinct from D00481 and X55881.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to make a transformed *Candida maltosa* according to claims 10-13, and 20-22 since the rationale, reagents, and process steps for making this transformed *Candida maltosa* are known. A transformed *Candida maltosa* according to claims 10-13, and 20-22 is expected to be

made by modifying the teachings of Kasuske *et al.* in which the genes taught by Takagi *et al.*, Takagi, Ohkuma, and Ohkuma *et al.* are cloned into the integrative vector taught by Kasuske *et al.* by methods well known in the art. This recombinant vector is then transformed into *Candida maltosa* by methods taught by Kasuske *et al.*

One of ordinary skill in the art would be motivated to make a transformed *Candida maltosa* according to claims 10-13, and 20-22 because of the advantage of higher levels of enzyme activity when multiple copies of a gene are integrated into the genome of the *Candida maltosa* strain. This advantage of higher levels of enzyme activity by integrating multiple copies of a gene into a host genome is well known in the art. Furthermore, one of ordinary skill in the art would have had a reasonable expectation for success of making a transformed *Candida maltosa* according to claims 10-13, and 20-22 because of the success of Kasuske *et al.* in transforming *Candida maltosa* with an integrative vector for this host cell.

Claims 8 and 9 are rejected under 35 U.S.C. 103(a) as being unpatentable over Hammond in view of Takagi *et al.*, Takagi, Ohkuma, Ohkuma *et al*, and Picataggio *et al.* (1992). Hammond teaches the bioproduction alkyl dicarboxylic acids from alkyl precursors using immobilized *Candida maltosa* cells (see entire patent). Hammond does not teach bioproduction alkyl dicarboxylic acids from alkyl precursors using a transformed *Candida maltosa* characterized by a genetically-engineered, enhanced alkane hydroxylating activity described by claim 8. The teaching of Takagi *et al.*, Takagi, Ohkuma, and Ohkuma *et al.* are described above. Picataggio *et al.* (1992) teach the disadvantages of using chemical synthesis to prepare aliphatic long-chain dicarboxylic acids which include the presence of byproducts during synthesis and extensive purification procedures to remove these byproducts (see first paragraph of the **Introduction**, p. 894).

It would have been obvious to one of ordinary skill in the art at the time the invention was made to enhance bioproduction of C_6 to C_{22} mono- and di-carboxylic acids according to claims 9 and 10 since the rationale, reagents, and process steps for this method are known. Enhancing bioproduction of of C_6 to C_{22} mono- and di-carboxylic acids according to claims 9 and 10 is expected by modifying the teachings of Hammond in the following manner:

A. Prepare the genetically-engineered *Candida maltosa* which is described above in the 35 U.S.C. 103(a) rejection of claims 10-13, and 20-22.

B. Use the genetically-engineered *Candida maltosa* in the process described by Hammond to produce any mono- or di-carboxylic acid.

One of ordinary skill in the art would be motivated to enhance bioproduction of C_6 to C_{22} mono- and di-carboxylic acids according to claims 9 and 10 since Picataggio *et al* (1992). teach the disadvantages of using chemical synthesis to prepare aliphatic long-chain dicarboxylic acids. In addition, the use and advantages of yeasts in the bioproduction of mono- and di-carboxylic

acids are well known in the art (for example see US 4, 275, 158 and US 4, 220, 720). Furthermore, one of ordinary skill in the art would have had a reasonable expectation for success in enhancing the bioproduction of mono- and dicarboylic acids according to claims 8 and 9 because of the success of Hammond in producing alkyl dicarboxylic acids from alkyl precursors.

Claims 25 and 26 are rejected under 35 U.S.C. 103(a) as being unpatentable over Kasuske *et al.* in view of Takagi *et al.*, Takagi, Ohkuma, Ohkuma *et al.*, Masuda *et al.* (1994), and Zimmer *et al.* The teachings of Kasuske *et al.* are described above. Kasuske *et al.* do not teach a DNA fragment according to claims 25 and 26. Takagi *et al.*, Takagi, Ohkuma, and Ohkuma *et al.* teach DNA sequences as described above which anticipate SEQ ID NOs: 35-43. Masuda *et al.* (1994) teach a *Candida maltosa* PGK promoter (see **Abstract**, p. 412; and ***Construction of an expression vector***, pp. 413-414). Zimmer *et al.* teach an efficient, reconstituted *Candida maltosa* monooxygenase system which was created by coexpressing *Candida maltosa* cytochrome P450 monooxygenase and cytochrome P450 reductase on a vector (see **Fig. 1**, p. 622). Furthermore, Zimmer *et al.* teach that this system is important because it allows for a simplified characterization of individual cytochrome P450 forms (see **Discussion**, p. 626).

It would have been obvious to one of ordinary skill in the art at the time the invention was made to make a DNA fragment according to claims 25 and 26 since the rationale, reagents, and process steps for making this DNA fragment are known. A DNA fragment according to claims 25 and 26 is expected to be made by modifying the teachings of Kasuske *et al.* as follows:

A. Prepare the first construct by ligating the PGK promoter taught by Masuda *et al.* (1994) to the DNA sequences encoding cytochrome P450 monooxygenase taught by Takagi *et al.*, Takagi, Ohkuma, and Ohkuma *et al.* by methods well know in the art.

B. Prepare the second construct by ligating the PGK promoter taught by Masuda *et al.* (1994) to the DNA sequences encoding cytochrome P450 reductase taught by Takagi *et al.*, Takagi, Ohkuma, and Ohkuma *et al.* by methods well known in the art.

C. Insert both constructs into the *Candida maltosa* vector taught by Kasuske *et al.* by methods well known in the art.

One of ordinary skill in the art would be motivated to make a DNA fragment according to claims 25 and 26 because Zimmer *et al.* teach that this combination of genes encoding cytochrome P450 monooxygenase and cytochrome P450 reductase on the same vector allows for a simplified characterization of individual cytochrome P450 forms. Furthermore, one of ordinary skill in the art would have had a reasonable expectation for success of making a DNA fragment according to claims 25 and 26 because the PGK promoter is known, the genes encoding cytochrome P450 monooxygenase and cytochrome P450 reductase are known, a *Candida*

maltosa vector is known, and methods for ligating the PGK promoter and these genes into a *Candida maltosa* vector are known.

Response to Arguments

5. Applicant's argument, filed January 4, 2000, have been fully considered and are addressed below.

Patentability Under 35 U.S.C.§ 112

Applicant has amended claims 4, 9, 11, 12, 20, and 26 by inserting Gene Bank references and sequence ID Nos. Rejection under 35 U.S.C. 112, first paragraph, has been withdrawn.

Patentability Under 35 U.S.C.§ 102

Applicant has amended claims 10 and 11 and presents arguments in view of these amended claims. However, claims 10 and 11 are now rejected under U.S.C. 103 as described above.

Applicant has amended claim 16 and presents arguments in view of the amended claim which are not persuasive. Claim 16 is now rejected under U.S.C. 102 as described above.

Applicant has canceled claim 24 in favor of claim 25, has amended claim 26, and presents arguments in view of these amended claims. However, claims 25 and 26 are now rejected under U.S.C. 103 as described above.

Patentability Under 35 U.S.C.§ 103

Applicant has amended claims 1, and 3-6 which are now rejected under U.S.C. 103 as described above. Claims 2 and 7 are allowed as described above.

Applicant has amended claims 20, 21, and 26 which are now rejected under U.S.C. 103 as described above. Claim 18 and 22 are allowed as described above.

Applicant presents arguments addressing the rejection of claims 8-23 and 27 under 35 U.S.C., 103 (a) on pp. 9-11 of the amendment, filed January 4, 2000, which are not persuasive. Masuda *et al* teach *Candida maltosa* mutant strains having no more than both POX4 genes disrupted (see **lines 15-24 of (c) Disruption of POX genes**, p159). In addition **Table 1** (see p. 159) shows that the P4DD strain which has both POX4 genes disrupted cannot use an alkane, alcohol, or a fatty-acid for growth. Therefore, the beta-oxidation pathway is blocked in strain P4DD. Because the beta-oxidation pathway has been blocked, it is inherent that production of dicarboxylic acids is increased. Using this *Candida maltosa* mutant strain in the process taught by Hammond as described above is expected to result in enhanced bioproduction of C_6 to C_{22} mono- and di-carboxylic acids. Accordingly, the rejection is maintained.

Conclusion

6. **THIS ACTION IS MADE FINAL.** Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the mailing date of this final action.

7. Claims 2, 7, 18, and 22 are allowed.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Christian L. Fronda whose telephone number is (703)305-1252. If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ponnathapura Achutamurthy, can be reached at (703)308-3804. The fax phone number for this Group is (703)308-0294. Any inquiry of a general nature or relating to the status of this application should be directed to the Group 1600 receptionist whose telephone number is (703)308-0196.

CLF
March 23, 2000

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